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PATENT TRADEMARK OFFICE

Docket No: 2094/1E286-US1

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re Application of: Jeffrey M. LINNEN and Kevin M. GORMAN

Serial No.: 09/493,353

Art Unit: 1634

Filed: January 28, 2000

Examiner: J. Goldberg

For: OLIGONUCLEOTIDE PRIMERS FOR EFFICIENT DETECTION OF HEPATITIS C  
VIRUS (HCV) AND METHODS OF USE THEREOF

COURTESY COPY OF PENDING CLAIMS  
(AS AMENDED APRIL 22, 2002)

1. (Twice Amended) A method for detecting the presence of Hepatitis C Virus (HCV) RNA in a biological sample, said method comprising:

- (A) performing a reverse transcription reaction using, as a template, RNA derived from said sample to produce HCV-specific reverse transcription products;

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- (B) amplifying said reverse-transcription products using one or more pairs of oligonucleotide primers specific for HCV to produce HCV-specific amplification products,

wherein said pairs are selected from the group consisting of:

- (a) forward primer 5'-CAGAAAGCGTCTAGCCATGGCGTTAGTA-3' (C69F28) <SEQ ID NO. 1> and reverse primer 5'-CGGTTCCGCAGAGACCACTATGGCTCTC-3' (C133R26) <SEQ ID NO. 4>; and
- (b) forward primer 5'-GGGAGAGCCATAGTGGTCTGCGGAA-3' (C131F25) <SEQ ID NO. 2> and reverse primer 5'-CGGGGCACTCGCAAGCACCCCTATCA-3' (C294R25) <SEQ ID NO. 7>; and

- (C) detecting said amplification products, wherein detection of said amplification products indicates the presence of HCV RNA in said sample.

2. A method as defined in claim 1, wherein said reverse transcription reaction is performed using random oligonucleotide primers.

3. A method as defined in claim 1, wherein said reverse transcription reaction is performed using one or more oligonucleotide primers having sequences corresponding to sequences in HCV RNA.

4. A method as defined in claim 1, wherein said amplifying is performed by a method selected from the group consisting of polymerase chain reaction, ligase chain reaction, strand displacement amplification, nucleic acid single base substitution, and transcription mediated amplification.

5. A method as defined in claim 1, wherein said detecting comprises visualizing said amplification products by gel electrophoresis.

6. A method as defined in claim 1, wherein said detecting comprises capturing said amplification products on a solid support containing one or more HCV-specific oligonucleotide probes and quantifying said captured products using a colorimetric assay.

7. A method as defined in claim 6, wherein said probes comprise a member selected from the group consisting of:

- (a) 5'-TTTCGCGACCCAACACTACTCGGCT-3' (C252-25-PRB) <SEQ ID NO. 13> and

- (b) 5'-CCTTCGCGACCCAACTACTCGGCT-3' (C252-27-PRB) <SEQ ID NO. 12> when said forward primer is (C131F25) or (C143F26); and

wherein said probes comprise

- (c) 5'-GGGTCCTGGAGGCTGCACGACACTCAT-3' (C96-22-PRB) <SEQ ID NO. 11> when said forward primer is (C69F28).

8. A method as defined in claim 1, wherein said sample is selected from the group consisting of blood, serum, plasma, urine, saliva, and cerebrospinal fluid.

9. (Twice Amended) A method for amplifying Hepatitis C Virus (HCV) DNA, which method comprises performing a polymerase chain reaction on a DNA sample containing HCV DNA using one or more pairs of oligonucleotide primers specific for HCV to produce HCV-specific amplification products, wherein said pairs are selected from the group consisting of:

- (a) forward primer 5'-CAGAAAGCGTCTAGCCATGGCGTTAGTA-3' (C69F28) <SEQ ID NO. 1> and reverse primer 5'-CGGTTCCGCGAGACCACTATGGCTCTC-3' (C133R26) <SEQ ID NO. 4>; and
- (b) forward primer 5'-GGGAGAGCCATAGTGGTCTGCGGAA-3' (C131F25) <SEQ ID NO. 2> and reverse primer

5'-CGGGGCACTCGCAAGCACCCCTATCA-3' (C294R25) <SEQ ID  
NO. 7>.

10. (Amended) A method as defined in claim 9, which method further comprises detecting said amplification products, wherein detection of said amplification products indicates the presence of HCV DNA in said sample.

11. A method as defined in claim 10, wherein said detecting comprises visualizing said amplification products by gel electrophoresis.

12. A method as defined in claim 10, wherein said detecting comprises capturing said amplification products on a solid support containing one or more HCV-specific oligonucleotide probes and quantifying said captured products using a colorimetric assay.

13. (Amended August 17, 2001) A method as defined in claim 12, wherein said probes comprise a member selected from the group consisting of:

(a) 5'-TTTCGGCGACCCAACACTACTCGGCT-3' (C252-25-PRB) <SEQ ID

NO. 13> and

(b) 5'-CCTTTCGGCGACCCAACACTACTCGGCT-3' (C252-27-PRB) <SEQ

ID NO. 12> when said forward primer is (C131F25) or (C143F26); and

wherein said probes comprise

- (c) 5'-GGGTCTCTGGAGGCTGCACGACACTCAT-3' (C96-22-PRB) <SEQ ID NO. 11> when said forward primer is (C69F28).

14. A method for detecting the presence of Hepatitis C Virus (HCV) RNA in a biological sample, said method comprising:

- (A) performing a reverse transcription reaction using as a template RNA derived from said sample to produce HCV-specific reverse transcription products;
- (B) amplifying said reverse-transcription products using a forward primer and a reverse primer to produce HCV-specific amplification products, wherein said forward primer consists of the oligonucleotide 5'-GGTGGCTCCATCTTAGCCCTAGTCACG-3' (1F27) <SEQ ID NO. 8> and said reverse primer consists of the oligonucleotide 5'-AGGCCAGTATCAGCACTCTCTGCAGTC-3' (57R27) <SEQ ID NO. 9>; and
- (C) detecting said amplification products,

wherein detection of said amplification products indicates the presence of HCV RNA in said sample.

15. A method as defined in claim 14, wherein said reverse transcription reaction is performed using random oligonucleotide primers.

16. A method as defined in claim 14, wherein said reverse transcription reaction is performed using one or more oligonucleotide primers having sequences corresponding to sequences in HCV RNA.

17. A method as defined in claim 14, wherein said amplifying is performed by a method selected from the group consisting of polymerase chain reaction, ligase chain reaction, strand displacement amplification, nucleic acid single base substitution, and transcription mediated amplification.

18. A method as defined in claim 14, wherein said detecting comprises visualizing said amplification products by gel electrophoresis.

19. A method as defined in claim 14, wherein said detecting comprises capturing said amplification products on a solid support containing one or more HCV-specific oligonucleotide probes and quantifying said captured products using a colorimetric assay.

20. A method as defined in claim 19, wherein said probes are selected from the group consisting of 5'-GCGGCTCACGGACCTTTCACAGCTA-3' (30PRB25) <SEQ ID NO. 14> and 5'-ATGCGGCTCACGGACCTTTCACAGC-3' (32PRB25) <SEQ ID NO. 15>.

21. A method as defined in claim 14, wherein said sample is selected from the group consisting of blood, serum, plasma, urine, saliva, and cerebrospinal fluid.

22. (Amended) A method for amplifying Hepatitis C Virus (HCV) DNA, which method comprises performing a polymerase chain reaction on a DNA sample containing HCV DNA using a forward primer and a reverse primer to produce HCV-specific amplification products, wherein said forward primer consists of the oligonucleotide 5'-GGTGGCTCCATCTTAGCCCTAGTCACG-3' (1F27) <SEQ ID NO. 8> and said reverse primer consists of the oligonucleotide 5'-AGGCCAGTATCAGCACTCTCTGCAGTC-3' (57R27) <SEQ ID NO. 9>.

23. (Amended) A method as defined in claim 22, which method further comprises detecting said amplification products, wherein detection of said amplification products indicates the presence of HCV DNA in said sample.

24. A method as defined in claim 23, wherein said detecting comprises visualizing said amplification products by gel electrophoresis.

25. A method as defined in claim 23, wherein said detecting comprises capturing said amplification products on a solid support containing one or more HCV-specific oligonucleotide probes and quantifying said captured products using a colorimetric assay.



26. A method as defined in claim 25, wherein said probes are selected from the group consisting of 5'-GCGGCTCACGGACCTTTCACAGCTA-3' (30PRB25) <SEQ ID NO. 14> and 5'-ATGCGGCTCACGGACCTTTCACAGC-3' 32PRB25) <SEQ ID NO. 15>.

27. (Twice Amended) A method for detecting the presence of Hepatitis C Virus (HCV) RNA in a biological sample, said method comprising:

- (A) performing a reverse transcription reaction using as a template RNA derived from said sample to produce HCV-specific reverse transcription products;
- (B) amplifying said reverse-transcription products using one or more pairs of 5' NCR oligonucleotide primers specific for HCV and one or more pairs of 3' NCR oligonucleotide primers to produce HCV-specific amplification products, wherein said 5' NCR primer pairs are selected from the group consisting of:
  - (a) forward primer 5'-CAGAAAGCGTCTAGCCATGGCGTTAGTA-3' (C69F28) <SEQ ID NO. 1> and reverse primer 5'-CGGTTCCGCAGACCACTATGGCTCTC-3' (C133R26) <SEQ ID NO. 4>; and
  - (b) forward primer 5'-GGGAGAGCCATAGTGGTCTGCGGAA-3' (C131F25) <SEQ ID NO. 2> and reverse primer 5'-CGGGGCACTCGCAAGCACCCCTATCA-3' (C294R25) <SEQ ID NO. 7>; and

wherein each of said pairs of 3' NCR oligonucleotide primers comprises a forward primer consisting of the oligonucleotide 5'-GGTGGCTCCATCTTAGCCCTAGTCACG-3' (1F27) <SEQ ID NO. 8> and a reverse primer consisting of the oligonucleotide 5'-AGGCCAGTATCAGCACTCTCTGCAGTC-3' (57R27) <SEQ ID NO. 9>; and

- (C) detecting said amplification products, wherein detection of said amplification products indicates the presence of HCV RNA in said sample.

28. A method as defined in claim 27, wherein said reverse transcription reaction is performed using random oligonucleotide primers.

29. A method as defined in claim 27, wherein said reverse transcription reaction is performed using one or more oligonucleotide primers having sequences corresponding to sequences in HCV RNA.

30. A method as defined in claim 27, wherein said amplifying is performed by a method selected from the group consisting of polymerase chain reaction, ligase chain reaction, strand displacement amplification, nucleic acid single base substitution, and transcription mediated amplification.

31. A method as defined in claim 27, wherein said detecting comprises visualizing said amplification products by gel electrophoresis.

32. A method as defined in claim 27, wherein said detecting comprises capturing said amplification products on a solid support containing one or more HCV-specific oligonucleotide probes and quantifying said captured products using a colorimetric assay.

33. A method as defined in claim 32, wherein said probes comprise a member selected from the group consisting of:

- (a) 5'-TTTCGCGACCCAACTACTCGGCT-3' (C252-25-PRB) <SEQ ID NO. 13> and
- (b) 5'-CCTTTCGCGACCCAACTACTCGGCT-3' (C252-27-PRB) <SEQ ID NO. 12> when said 5' NCR forward primer is (C131F25) or (C143F26);

wherein said probes comprise

- (c) 5'-GGGTCCTGGAGGCTGCACGACACTCAT-3' (C98-22-PRB) <SEQ ID NO. 11> when said 5' NCR forward primer is (C69F28); and

wherein said probes comprise a member selected from the group consisting of

- (d) 5'-GCGGCTCACGGACCTTTCACAGCTA-3' (30PRB25) <SEQ ID NO. 14>; and

- (e) 5'-ATGCGGCTCACGGACCTTTCACAGC-3' (32PRB25) <SEQ ID NO. 15>.

34. A method as defined in claim 27, wherein said sample is selected from the group consisting of blood, serum, plasma, urine, saliva, and cerebrospinal fluid.

35. (Twice Amended) A method for amplifying Hepatitis C Virus (HCV) DNA, which method comprises performing a polymerase chain reaction on a DNA sample containing HCV DNA using one or more pairs of 5' NCR oligonucleotide primers specific for HCV and one or more pairs of 3' NCR oligonucleotide primers to produce HCV-specific amplification products, wherein said 5' NCR primer pairs are selected from the group consisting of:

- (a) forward primer 5'-CAGAAAGCGTCTAGCCATGGCGTTAGTA-3' (C69F28) <SEQ ID NO. 1> and reverse primer 5'-CGGTTCCGCAGACCACTATGGCTCTC-3' (C133R26) <SEQ ID NO. 4>; and
- (b) forward primer 5'-GGGAGAGCCATAGTGGTCTGCGGAA-3' (C131F25) <SEQ ID NO. 2> and reverse primer 5'-CGGGGCACTCGCAAGCACCCATCA-3' (C294R25) <SEQ ID NO. 7>; and

wherein each of said pairs of 3' NCR oligonucleotide primers comprises a forward primer consisting of the oligonucleotide 5'-GGTGGCTCCATCTTAGCCCTAGTCACG-3' (1F27)

<SEQ ID NO. 8> and a reverse primer consisting of the oligonucleotide 5'-AGGCCAGTATCAGCACTCTCTGCAGTC-3' (57R27) <SEQ ID NO. 9>.

36. (Amended) A method as defined in claim 35, which method further comprises detecting said amplification products, wherein detection of said amplification products indicates the presence of HCV DNA in said sample.

37. A method as defined in claim 36, wherein said detecting comprises visualizing said amplification products by gel electrophoresis.

38. A method as defined in claim 36, wherein said detecting comprises capturing said amplification products on a solid support containing one or more HCV-specific oligonucleotide probes and quantifying said captured products using a colorimetric assay.

39. A method as defined in claim 38, wherein said probes comprise a member selected from the group consisting of:

- (a) 5'-TTTCGCGACCCAACTACTCGGCT-3' (C252-25-PRB) <SEQ ID NO. 13> and
- (b) 5'-CCTTTCGCGACCCAACTACTCGGCT-3' (C252-27-PRB) <SEQ ID NO. 12> when said 5' NCR forward primer is (C131F25) or (C143F26);

wherein said probes comprise

- (c) 5'-GGGTCTGGAGGCTGCACGACACTCAT-3' (C96-22-PRB) <SEQ ID NO. 11> when said 5' NCR forward primer is (C69F28); and

wherein said probes comprise a member selected from the group consisting of

- (d) 5'-GCGGCTCACGGACCTTTCACAGCTA-3' (30PRB25) <SEQ ID NO. 14>; and  
(e) 5'-ATGCGGCTCACGGACCTTTCACAGC-3' (32PRB25) <SEQ ID NO. 15>.

40. (Twice Amended) An oligonucleotide selected from the group consisting of:

- 5'-CAGAAAGCGTCTAGCCATGGCGTTAGTA-3' (C69F28) <SEQ ID NO. 1>;  
5'-GGGAGAGCCATAGTGGTCTGCGGAA-3' (C131F25) <SEQ ID NO. 2>;  
5'-CGGTTCCGAGACCACTATGGCTCTC-3' (C133R26) <SEQ ID NO. 4>;  
5'-CGGGGCACTCGCAAGCACCCCTATCA-3' (C294R25) <SEQ ID NO. 7>;  
5'-GGTGGCTCCATCTTAGCCCTAGTCAGC-3' (1F27) <SEQ ID NO. 8>;  
5'-AGGCCAGTATCAGCACTCTCTGCAGTC-3' (57R27) <SEQ ID NO. 9>;  
5'-GGGTCTGGAGGCTGCACGACACTCAT-3' (C96-22-PRB) <SEQ ID NO. 11>;  
5'-CCTTTCGCGACCCAACTACTCGGCT-3' (C252-27-PRB) <SEQ ID NO. 12>;  
5'-TTTCGCGACCCAACTACTCGGCT-3' (C252-25-PRB) <SEQ ID NO. 13>;  
5'-GCGGCTCACGGACCTTTCACAGCTA-3' (30PRB25) <SEQ ID NO. 14>; and  
5'-ATGCGGCTCACGGACCTTTCACAGC-3' (32PRB25) <SEQ ID NO. 15>.

41. (Twice Amended) An HCV-specific amplification primer oligonucleotide selected from the group consisting of:

5'-CAGAAAGCGTCTAGCCATGGCGTTAGTA-3' (C69F28) <SEQ ID NO. 1>;

5'-GGGAGAGCCATAGTGGTCTGCGGAA-3' (C131F25) <SEQ ID NO. 2>;

5'-CGGTTCCGCAGACCACTATGGCTCTC-3' (C133R26) <SEQ ID NO. 4>;

5'-CGGGGCACTCGCAAGCACCTATCA-3' (C294R25) <SEQ ID NO. 7>;

5'-GGTGGCTCCATCTTAGCCCTAGTCACG-3' (1F27) <SEQ ID NO. 8>; and

5'-AGGCCAGTATCAGCACTCTCTGCAGTC-3' (57R27) <SEQ ID NO. 9>.

42. (Twice amended) An oligonucleotide probe selected from the group consisting of:

5'-GGGTCCTGGAGGCTGCACGACACTCAT-3' (C96-22-PRB) <SEQ ID NO. 11>;

5'-CCTTTCGCGACCCAACACTACTCGGCT-3' (C252-27-PRB) <SEQ ID NO. 12>;

5'-TTTCGCGACCCAACACTACTCGGCT-3' (C252-25-PRB) <SEQ ID NO. 13>;

5'-GCGGCTCACGGACCTTTCACAGCTA-3' (30PRB25) <SEQ ID NO. 14>; and

5'-ATGCGGCTCACGGACCTTTCACAGC-3' (32PRB25) <SEQ ID NO. 15>.

43. (Amended) A kit for amplifying HCV DNA derived from HCV RNA, said kit comprising one or more pairs of 5' NCR oligonucleotide primers, wherein said 5' NCR primer pairs are selected from the group consisting of:

- (a) forward primer 5'-CAGAAAGCGTCTAGCCATGGCGTTAGTA-3' (C69F28) <SEQ ID NO. 1> and reverse primer 5'-CGGTTCCGCAGACCACTATGGCTCTC-3' (C133R26) <SEQ ID NO. 4>; and
- (b) forward primer 5'-GGGAGAGCCATAGTGGTCTGCGGAA-3' (C131F25) <SEQ ID NO. 2> and reverse primer 5'-CGGGGCACTCGCAAGCACCTATCA-3' (C294R25) <SEQ ID NO. 7>.

44. A kit as defined in claim 43, further comprising one or more pairs of 3' NCR oligonucleotide primers, wherein each of said pairs of 3' NCR oligonucleotide primers comprises a forward primer consisting of the oligonucleotide 5'-GGTGGCTCCATCTTAGCCCTAGTCACG-3' (1F27) <SEQ ID NO. 8> and a reverse primer consisting of the oligonucleotide 5'-AGGCCAGTATCAGCACTCTCTGCAGTC-3 (57R27) <SEQ ID NO. 9>.

45. A kit as defined in claim 43, further comprising one or more probes.

46. A kit as defined in claim 44, further comprising one or more probes.



47. A kit as defined in claim 45, wherein said probes comprise a member selected from the group consisting of:

- (a) 5'-TTTCGCGACCCAACACTACTCGGCT-3' (C252-25-PRB) <SEQ ID NO. 13> and
- (b) 5'-CCTTCGCGACCCAACACTACTCGGCT-3' (C252-27-PRB) <SEQ ID NO. 12> when said 5' NCR forward primer is (C131F25) or (C143F26); and

wherein said probes comprise

- (c) 5'-GGGTCCTGGAGGCTGCACGACACTCAT-3' (C96-22-PRB) <SEQ ID NO. 11> when said 5' NCR forward primer is (C69F28).

48. A kit as defined in claim 46, wherein said probes comprise a member selected from the group consisting of:

- (a) 5'-TTTCGCGACCCAACACTACTCGGCT-3' (C252-25-PRB) <SEQ ID NO. 13> and
- (b) 5'-CCTTCGCGACCCAACACTACTCGGCT-3' (C252-27-PRB) <SEQ ID NO. 12> when said 5' NCR forward primer is (C131F25) or (C143F26);

wherein said probes comprise

- (c) 5'-GGGTCCTGGAGGCTGCACGACACTCAT-3' (C96-22-PRB) <SEQ ID NO. 11> when said 5' NCR forward primer is (C69F28); and

wherein said probes comprise a member selected from the group consisting of

- (d) 5'-GCGGCTCACGGACCTTTCACAGCTA-3' (30PRB25) <SEQ ID NO. 14>; and
- (e) 5'-ATGCGGCTCACGGACCTTTCACAGC-3' (32PRB25) <SEQ ID NO. 15>.

49. A kit as defined in claim 43, wherein said pair of 5' NCR primers consists of 5'-CAGAAAGCGTCTAGCCATGGCGTTAGTA-3' (C69F28) <SEQ ID NO. 1> and 5'-CGGTTCCGCAGACCACTATGGCTCTC-3' (C133R26) <SEQ ID NO. 4>.

50. A kit as defined in claim 43, wherein said pair of 5' NCR primers consists of 5'-GGGAGAGCCATAGTGGTCTGCGGAA-3' (C131F25) <SEQ ID NO. 2> and 5'-CGGGGCACTCGCAAGCACCCCTATCA-3' (C294R25) <SEQ ID NO. 7>.

51. A kit for amplifying HCV cDNA derived from HCV RNA, said kit comprising one or more pairs of 3' NCR oligonucleotide primers, wherein each of said pairs of 3' NCR oligonucleotide primers comprises a forward primer consisting of the oligonucleotide 5'-GGTGGCTCCATCTTAGCCCTAGTCAG-3' (1F27) <SEQ ID NO. 8> and a reverse primer consisting of the oligonucleotide 5'-AGGCCAGTATCAGCACTCTCTGCAGTC-[3] 3' (57R27) <SEQ ID NO. 9>.

52. A kit as defined in claim 51, further comprising one or more probes.

53. A kit as defined in claim 52, wherein said probes are selected from the group consisting of:

- (a) 5'-GCGGCTCACGGACCTTTCACAGCTA-3' (30PRB25) <SEQ ID NO. 14>;  
and
- (b) 5'-ATGCGGCTCACGGACCTTTCACAGC-3' (32PRB25) <SEQ ID NO. 15>.

54. (Twice Amended) A kit for detecting the presence of HCV DNA, said kit comprising one or more pairs of 5' NCR oligonucleotide primers, wherein said 5' NCR primer pairs are selected from the group consisting of:

- (a) forward primer 5'-CAGAAAGCGTCTAGCCATGGCGTTAGTA-3' (C69F28) <SEQ ID NO. 1> and reverse primer 5'-CGGTTCCGCAGACCACTATGGCTCTC-3' (C133R26) <SEQ ID NO. 4>; and
- (b) forward primer 5'-GGGAGAGCCATAGTGGTCTGCGGAA-3' (C131F25) <SEQ ID NO. 2> and reverse primer 5'-CGGGGCACTCGCAAGCACCCTATCA-3' (C294R25) <SEQ ID NO. 7>.

55. A kit as defined in claim 54, further comprising one or more pairs of 3' NCR oligonucleotide primers, wherein each of said pairs of 3' NCR oligonucleotide primers comprises a forward primer consisting of the oligonucleotide 5'-GGTGGCTCCATCTTAGCCCTAGTCACG-3' (1F27) <SEQ ID NO. 8> and a reverse primer consisting of the oligonucleotide 5'-AGGCCAGTATCAGCACTCTCTGCAGTC-3' (57R27) <SEQ ID NO. 9>.

56. A kit as defined in claim 54, further comprising one or more probes.

57. A kit as defined in claim 55, further comprising one or more probes.

58. A kit as defined in claim 56, wherein said probes comprise a member selected from the group consisting of:

- (a) 5'-TTTCGCGACCCAACACTACTACTCGGCT- 3' (C252-25-PRB) <SEQ ID NO. 13> and
- (b) 5'-CCTTTTCGCGACCCAACACTACTCGGCT- 3' (C252-27-PRB) <SEQ ID NO. 12> when said 5' NCR forward primer is (C131F25) or (C143F26);

wherein said probes comprise

- (c) 5'-GGGTCCTGGAGGCTGCACGACACTCAT-3' (C96-22-PRB) <SEQ ID NO. 11> when said 5' NCR forward primer is (C69F28).

59. A kit as defined in claim 57, wherein said probes comprise a member selected from the group consisting of:

- (a) 5'-TTTCGCGACCCAACACTACTCGGCT-3' (C252-25-PRB) <SEQ ID NO. 13> and
- (b) 5'-CCTTTCGCGACCCAACACTACTCGGCT-3' (C252-27-PRB) <SEQ ID NO. 12> when said 5' NCR forward primer is (C131F25) or (C143F26);

wherein said probes comprise

- (c) 5'-GGGTCCTGGAGGCTGCACGACACTCAT-3' (C96-22-PRB) <SEQ ID NO. 11> when said 5' NCR forward primer is (C69F28); and

wherein said probes comprise a member selected from the group consisting of

- (d) 5'-GCGGCTCACGGACCTTTCACAGCTA-3' (30PRB25) <SEQ ID NO. 14>; and
- (e) 5'-ATGCGGCTCACGGACCTTTCACAGC-3' (32PRB25) <SEQ ID NO. 15>.

60. A kit as defined in claim 54, wherein said pair of 5' NCR primers consists of 5'-CAGAAAGCGTCTAGCCATGGCGTTAGTA-3' (C69F28) <SEQ ID NO. 1> and 5'-CGGTTCCGCAGACCACTATGGCTCTC-3' (C133R26) <SEQ ID NO. 4>.

61. A kit as defined in claim 54, wherein said pair of 5' NCR primers consists of 5'-GGGAGAGCCATAGTGGTCTGCGGAA- 3' (C131F25) <SEQ ID NO. 2> and 5'-CGGGGCACTCGCAAGCACCTATCA- 3' (C294R25) <SEQ ID NO. 7>.

62. A kit for detecting the presence of HCV RNA, said kit comprising one or more pairs of 3' NCR oligonucleotide primers, wherein each of said pairs of 3' NCR oligonucleotide primers comprises a forward primer consisting of the oligonucleotide 5'-GGTGGCTCCATCTTAGCCCTAGTCACG-3' (1F27) <SEQ ID NO. 8> and a reverse primer consisting of the oligonucleotide 5'-AGGCCAGTATCAGCACTCTCTGCAGTC-3 (57R27) <SEQ ID NO. 9>.

63. A kit as defined in claim 62, further comprising one or more probes.

64. A kit as defined in claim 63, wherein said probes are selected from the group consisting of 5'-GCGGCTCACGGACCTTTCACAGCTA-3' (30PRB25) <SEQ ID NO. 14> and 5'-ATGCGGCTCACGGACCTTTCACAGC-3' (32PRB25) <SEQ ID NO. 15>.